BIOLOGICAL CHARACTERISTICS OF UNA VIRUS

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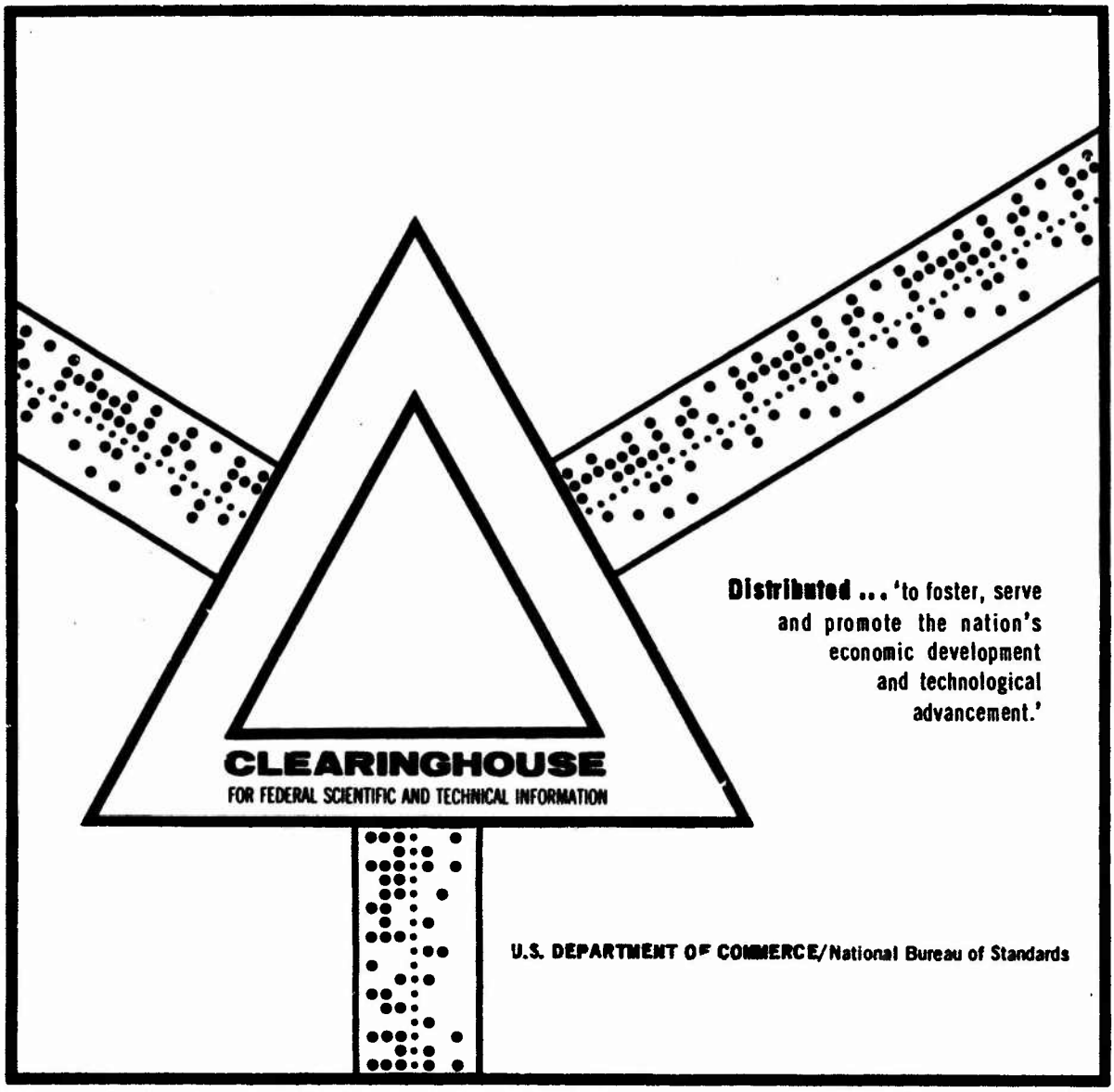

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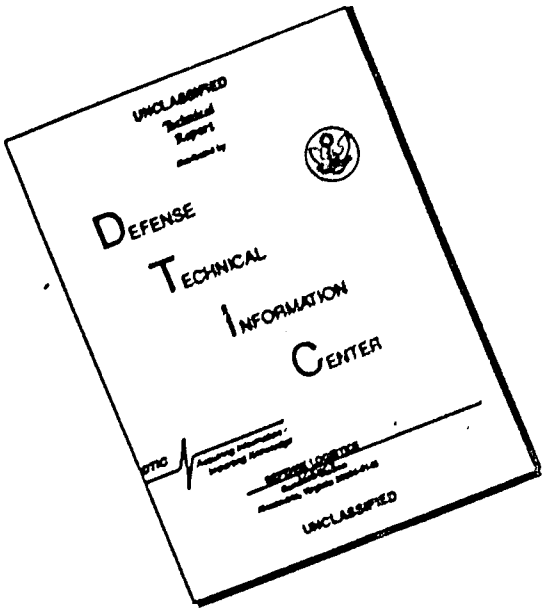

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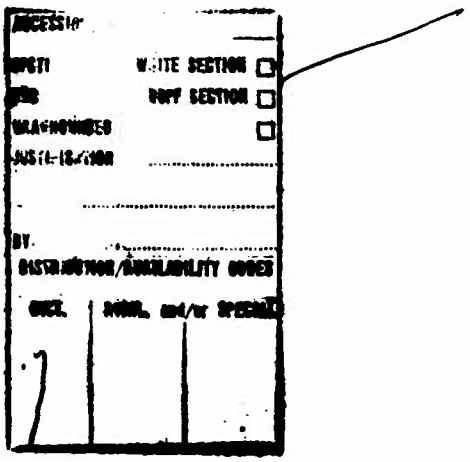
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by

O. N. Shcheglovitova

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BIOLOGICAL CHARACTERISTICS OF UNA VIRUS
(Report 2)

Ivanovskiy Institute of Virology O.N. Shcheglovitova
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Interest in arbor viruses has heightened recently in view of the isolation of such viruses in many of the world's countries. The present investigation deals with the biological distinctions and development of optimum methods for passing and preserving Una arbor virus which is referable to group A according to the Casals classification. In the course of this study it was found that Una virus induces CPE [cytopathogenic effect] in VNK-21 tissue cultures and plaque production in chick fibroblast cultures. The clinical symptoms of pathology following infection of albino mice weighing 6-7 grams with Una virus, intracerebrally, are characterized by acute development of disease, paralysis of the hind legs on the 4th-5th postinfection day. The virus is demonstrable in the blood in the highest titers 6 and 48 hours after infection, and in the brain 72 hours after infection. Una virus can be preserved by means of lyophilization of a brain suspension with and without fillers in a vacuum dryer. Of those we studied 5% peptone was the best filler.[stabilizer]

The heightened recent interest in investigation of arbor viruses is related to their isolation in many of the nations of the world [1, 5]. We made a study of the biological distinctions and developed optimum methods of passing and preserving Una arbor virus which is referable to group A arborviruses according to the Casals classification [6]. There is no detailed description of the properties of this virus in the literature.

the cellular layer. After maintaining the culture at 33° for four days thesefindings do not change. On the VNK-21 culture the CPD_{50} constitutes $10^{-6,6}$ - $10^{-6,6}$. The PP dosage on the CE culture constitutes 5·10 PPU/ml [plaque producing units per milliliter].

Table 1

CPE and PP of Una virus in different tissue cultures

(a) Культура ткани	(b) Цитопатический эффект	Бляшкообразующая способность вируса (c)	
		метод Портерфилда (d)	метод Карповича (e)
ВНК-21 VNK-21	+	—[1]	0
КЭ CE	—	+	+
ЭЧ HEK	—	0	0
HEp-2	—	0	0
СМ CMH	—	0	0

Legend:

a) tissue culture
b) CPE
c) PP capacity of the virus
d) Porterfield method
e) Karpovich method

[1]The VNK-21 tissue culture is not suitable for demonstrating the plaque producing capacity of the virus since the cells degenerate within 24 hours under an agar cover.

Una virus was able to form plaques only on the CE culture. They were visible on the second postinfection day. Their number and appearance did not change upon further cultivation at 33°. The plaques are round, 1-1.5 mm in diameter, with irregular borders. The appearance of the plaques formed on CE is the same when using different covers, but they are less transparent when covered by the Karpovich method.

Table 2 shows that the virus appeared in the blood of mice only six hours after inoculation. Una virus is characterized by two rises in blood titers: six ($10^{-2,2}$) and 48 ($10^{-2,3}$) hours after infection. The virus was demonstrable in brain tissue three hours after inoculation and the quantity increased gradually. The highest titer was observed after 72 hours ($10^{-4,3}$), thereafter it declined gradually. On the seventh day the titer was 10^{-1}.

In observing the sick mice we noticed that they were much less active than the healthy ones, their fur was often ruffled, and paralysis of the hind extremities was observed on the 4th-5th postinoculation day. Most of the animals died on the 4th day after infection.

Table 2
Dynamics of accumulation of Una virus in the blood and brain tissue of infected mice

(a) Источник вируса	(b) [illegible]								
	3	6	[illegible]	[illegible]	[illegible]	72	[illegible]	142	166
Мозг (c)	0,6*	1,6	1,7	[illegible]	[illegible]	4,3	3,7	2,2	1
Кровь (d)	0	2,2	1,3	2	2,3	0,9	1,2	0,5	0,5

Legend:
a) source of virus
b) time after infection (hours)
c) brain
d) blood

Note: the numbers in the table indicate the lg of viral titers

Table 3
Changes in viral titers after desiccation and storage of lyophilized suspensions at different temperatures

(a) Способ консервации	(b) До сушки	(c) После сушки	(d) Срок хранения									
			(e) 1 месяц				2 месяца		3 месяца		7 месяцев	
			(f) способ хранения									
			Т	К	Ф	Z	Т	К	Т	К	Ф	Z
(g) Вирус на физиологическом растворе без наполнителя	4,6	5,1	1,7	1	3,5	3	1	1	1	1,7	2,6	3
(h) Вирус + 10% сахарозы + 1% желатины	4,3	4,7	2,8	3,6	2,5	3,7	1	3	1	2,7	3	4,1
(i) Вирус + 20% молока	4,6	5,4	4	6	4,5	4,3	3,8	4,3	1	3,5	4,4	3,1
(j) Вирус + 5% пептона	4,2	5	3,5	5,5	4,3	4,6	2	4,3	2	4,8	4,1	4,3

Legend:
a) means of preservation
b) before desiccation
c) after "
d) storage time
e) ... months
f) storage method
g) virus on saline without stabilizer
h) " + 10% saccharose + 1% gelatin
i) " + 20% milk
j) " + 5% peptone

Note: the numbers in the table indicate the lg of viral titers

Table 3 shows the changes in viral titers in the course of drying and storage of lyophilized suspensions at different temperatures. As we see the virus containing suspension both with and without stabilizers tolerates the drying process well under the conditions chosen. A marked drop in titers was noted when Una virus, dried without stabilizers, was stored at thermal and room temperature for one month. The optimum stabilizer is peptone in an end concentration of 5%. It as well as 20% milk retar-ed the drop in viral titer while stored at 4° for the duration of the observation period. Virus stored at -20° without stabilizer and with stabilizers also retained its titers well during the observation period.

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13. ABSTRACT

Interest in arbor viruses has heightened recently in view of the isolation of such viruses in many of the world's countries. The present investigation deals with the biological distinctions and development of optimum methods for passing and preserving UNA arbor virus which is referable to group A according to the Casals classification. In the course of this study it was found that UNA virus induces CPE in VNK-21 tissue cultures and plaque production in chick fibroblast cultures. The clinical symptoms of pathology following infection of albino mice weighing 6-7 grams with UNA virus, intracerebrally, are characterized by acute development of disease, paralysis of the hind legs on the 4th-5th postinfection day. The virus is demonstrable in the blood in the highest titers 6 and 48 hours after infection, and in the brain 72 hours after infection. UNA virus can be preserved by means of lyophilization of a brain suspension with and without fillers in a vacuum dryer. Of those we studied 5% peptone was the best filler (stabilizer).

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VNK-21 tissue cultures						
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Fibroblast cultures						
Lyophilization						